

Research Article

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Evidence for the proposal that *Palisada corallopsis* and *P. furcata* (Ceramiales, Rhodophyta) are conspecific

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Abstract: *Palisada corallopsis* has a widely reported distribution in the western and eastern Atlantic and some areas of the Indo-Pacific Ocean, whereas *P. furcata* has a more restricted distribution, reported only from Brazil and Cuba. *Palisada corallopsis* and *P. furcata* were collected from their type localities, Havana, Cuba and Ceará, Brazil, respectively, and compared by morphological and molecular analyses using *rbcL* and COI-5P genes. The molecular results placed the sequences from the type locality of both species in the same clade, showing low genetic divergence, 0.95 % for *rbcL* and 1.6 % for COI-5P. Morphologically, the species share similar habits, with branchlets bi-to trifurcated, cortical cells that may be arranged as a palisade and connected by secondary pit-connections. Our detailed morphological observations, including examination of the type specimens, and molecular analyses demonstrate that

P. corallopsis and *P. furcata* are conspecific, *P. furcata* being a later taxonomic synonym of *P. corallopsis*.

Keywords: COI-5P; morphology; *Palisada*; *rbcL*; taxonomy

1 Introduction

The *Laurencia* complex currently comprises 217 taxonomically accepted species (Guiry and Guiry 2024) assigned to eight genera: *Laurencia sensu stricto* (Lamouroux 1813), *Osmundea* (Nam et al. 1994; Stackhouse 1809), *Chondrophycus* (Garbary and Harper 1998), *Palisada* Nam (2007), *Yuzurua* (Martin-Lescanne et al. 2010), *Laurenciella* (Cassano et al. 2012a), *Ohelopapa* (Rousseau et al. 2017), and *Corynecladia* (as *Coronaphycus*) (Cassano et al. 2019; Metti et al. 2015). *Palisada* is the second largest genus of the *Laurencia* complex with 26 accepted species names and is distributed from tropical to subtropical regions of the Atlantic and Indo-Pacific Oceans (Guiry and Guiry 2024).

Historically, the genus *Laurencia* was split into two subgenera: *Laurencia* and *Chondrophycus* Tokida et Saito; the latter having two sections, section *Chondrophycus* and section *Palisadae* Yamada (Saito 1967). The subgenus *Chondrophycus* was raised to the generic rank by Garbary and Harper (1998). Based on this new delineation, Nam (1999) proposed a new infrageneric circumscription for the genus *Chondrophycus* splitting it into four subgenera: *Chondrophycus*, *Kangjaewonia* K.W. Nam, *Palisada* (Yamada) K.W. Nam and *Yuzurua* K.W. Nam. Within the subgenus *Palisada*, Nam (1999) accommodated two sections: *Palisadae* and *Papilosae* (J. Agardh) K.W. Nam. Later, Nam (2006) made a new generic delineation of the *Laurencia* complex with four major clades recognized: *Laurencia*, the *Chondrophycus palisadus* group, the *C. cartilagineus* group and *Osmundea*. Nam's (2006) results showed that the genus *Chondrophycus* was paraphyletic and that the *C. palisadus* group should be treated as an independent genus. Thus, Nam (2006) proposed the genus *Palisada* based on Yamada's (1931) section *Palisadae*, with *Palisada robusta nomen novum* as the

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type species, renamed to avoid a tautonym. The name *Palisada* and the new combinations proposed for *Palisada* species by Nam (2006) were validated in a later publication (Nam 2007) due to the lack of Latin diagnosis in the previous publication (Nam 2006).

Based on the type species, *Palisada robusta* K.W. Nam, the genus is characterized by a combination of vegetative and reproductive features such as: (i) vegetative axial segments with two pericentral cells, (ii) first pericentral cell produced underneath the basal cell of the trichoblast, (iii) a palisade arrangement of the cortical layer in some species, (iv) spermatangial development of the trichoblast type, (v) spermatangial branches produced from one of two laterals on a suprabasal cell of the trichoblasts, (vi) procarp-bearing segments with four pericentral cells, (vii) tetrasporangia arranged at right angles to the main axis, (viii) tetrasporangia originated from particular pericentral cells (the second pericentral cell is always fertile, the first one remaining sterile), (ix) additional fertile pericentral cells may be produced in the tetrasporangial axis (Nam 2006, 2007; Nam et al. 1994). Procarp-bearing segments with five pericentral cells were described for *Palisada poiteaui* (J.V. Lamouroux) K.W. Nam and its var. *gemmifera* (Harvey) M.J. Wynne (Fujii et al. 1996). Later, *P. poiteaui* was transferred to *Yuzurua*, *Y. poiteaui* (J.V. Lamouroux) Martin-Lescanne in Martin-Lescanne et al. (2010) and *Yuzurua poiteaui* var. *gemmifera* (Wynne 2011).

Molecular studies carried out on *Palisada* species from the western and eastern Atlantic inferred phylogenetic affinities, proposed the conspecificity of species and proposed new combinations (Cassano et al. 2009, 2012b; Collado-Vides et al. 2017; Fujii et al. 2006; Popolizio et al. 2022; Senties and Díaz-Larrea 2008). Of the 26 species described for the genus, six are cited for the western Atlantic: *Palisada cervicornis*, *P. corallopsis*, *P. flagellifera*, *P. furcata*, *P. intermedia* and *P. perforata* (Wynne 2022). Among these, *P. corallopsis* and *P. furcata* are the most closely related in terms of gross morphology and anatomical characteristics of the thallus (Cordeiro-Marino et al. 1994; Montagne 1842).

Palisada corallopsis is widely distributed in the western Atlantic Ocean from North Carolina, USA, to Brazil, including several Caribbean islands and Bermuda, whereas in the eastern Atlantic it is reported only around offshore islands in the Canary Islands, Madeira, Salvage Islands and Cape Verde Islands (Collado-Vides et al. 2017; Delnatte and Wynne 2016; Duncan and Lee Lum 2006; Herrera-Moreno and Betancourt Fernández 2022; John et al. 2004; Littler and Littler 1997; Neto et al. 2001; Popolizio et al. 2022; Schneider and Searles 1991; Schnetter 1969; Senties and Díaz-Larrea 2008; Suárez et al.

2023; Taylor 1960). Its distribution also extends to the Indo-Pacific Ocean but with a more restricted occurrence, reported only for Indonesia (Atmadja and Prud'homme van Reine 2012; Silva et al. 1996) and the Philippines (Ang et al. 2014; Gazon-Fortes 2012; Lastimoso and Santiañez 2021; Phang, S.-M. et al. 2016; Silva et al. 1987). On the other hand, *P. furcata* has a much more restricted distribution in the western Atlantic, occurring from Ceará (northeastern Brazil) to Espírito Santo (southeastern Brazil) (Cassano et al. 2012b; Cordeiro-Marino et al. 1994; Fujii and Senties 2005) and northeastern Cuba at 3 m depth in a *Thalassia* biotope (Areces et al. 2003; Suárez et al. 2023).

The molecular study of type specimens or specimens from the type locality is important in order to correctly apply specific names (Soares et al. 2019), and this can lead to the correction of misapplied names and synonymizations, representing a major advance in solving taxonomic problems. *Palisada corallopsis* and *P. furcata* are morphologically closely related and lack DNA sequences from their type localities. We collected samples of *P. corallopsis* from Havana, Cuba, and *P. furcata* from Ceará, Brazil corresponding to their type localities, and we performed molecular analyses using *rbcL* and COI-5P genes. We also compared these freshly collected materials with the holotypes of *P. corallopsis* and *P. furcata*, and provide for the first time a detailed and comparative morpho-anatomical study of both species.

2 Materials and methods

2.1 Morphology

Samples of *Palisada corallopsis* (Montagne) Senties, Fujii et Díaz-Larrea from Havana, Cuba, and *P. furcata* (Cordeiro-Marino et M.T.Fujii) Cassano et M.T.Fujii from Ceará state, Brazil, were collected between 2018 and 2020. Algal samples for morphological study were pressed as herbarium sheets, whereas material used in the molecular studies was dehydrated on silica gel. Transverse and longitudinal hand sections were made with a stainless-steel razor blade and stained with 0.5 % aqueous aniline blue solution, acidified with 1N HCl. Nine specimens were analyzed, and for each specimen studied, a minimum of 20 measurements of each morphometric character were made, when possible. Measurements are given as length × diameter. Photographs were taken with a Panasonic Lumix DMC-FH4 camera (Panasonic Corporation, Osaka, Japan) coupled to a Zeiss Stemi 2000-C stereomicroscope and Carl Zeiss Axio Scope A1 microscopes (Zeiss, Göttingen, Germany). Voucher

specimens are deposited in the Herbarium SP of the Environmental Research Institute, São Paulo, Brazil, and the National Aquarium of Cuba (HANC) collections. Additional specimens from SP and HANC herbaria, including the type material, were studied. Herbaria abbreviations follow the Index Herbariorum (Thiers 2024).

2.2 Molecular analysis

Total genomic DNA was extracted from silica gel-dried material after automated grinding in a Precellys[®]24 tissue homogenizer (Bertin Instruments, Montigny-le-Bretonneux, France) using the NucleoSpin[®] Plant II Kit (Macherey-Nagel, Düren, Germany) following the manufacturer's instructions. DNA sequences were generated for the plastid-encoded large subunit of the ribulose-1,5-bisphosphate carboxylase/oxygenase gene (*rbcL*) and the 5' region of the mitochondrial cytochrome *c* oxidase 1 gene (COI-5P) for *P. corallopsis* and *P. furcata*. Polymerase chain reactions (PCRs) were run in 25 µl volumes using GoTaq[®] Green Master Mix (Promega, Madison, Wisconsin, USA) in a Techne TC-512 thermocycler (Bibby Scientific Ltd, Staffordshire, UK). The *rbcL* gene was amplified in three fragments using specific primer pairs for *P. corallopsis* (Fstart-R492, F492-R1150, F993-*rbcL*revNew), and for *P. furcata* (F57-R753, F577-R1150, F993-R*rbcS*) (Freshwater and Rueness 1994; Saunders and Moore 2013). PCR conditions for *rbcL* followed Geraldino et al. (2009). The COI-5P region was amplified using the primers GazF1 and GazR1 (Saunders 2005), and PCR conditions described in Cassano et al. (2019) were used. Amplified products were checked for length, purity and yield in 1% agarose gels and purified with the Illustra GFX PCR DNA and Gel Band Purification Kit (GE Healthcare, Buckinghamshire, UK) following the manufacturer's recommendations. Successfully purified PCR products were sequenced using the Big Dye Terminator v.3.1 reaction cycle sequencing Kit (Applied Biosystems, Foster City, California, USA) on a 3,730 Genetic Analyzer (Applied Biosystems). Sequences were obtained by the sequencing service of the Human Genome Research Center (Biosciences Institute, University of São Paulo, Brazil).

2.3 Phylogenetic analysis

Forward and reverse reads were manually assembled to generate consensus sequences for each marker using Bio-Edit 7.2.6.1 (Hall 1999). The new sequences generated for *P. corallopsis* and *P. furcata* and additional sequences

retrieved from GenBank (Supplementary Table S1) were added to each dataset to construct the final alignments. *Chondria collinsiana* M. Howe (GU330225) and *Chondria dasyphylla* (Woodward) C. Agardh (U04021) were used as outgroups for *rbcL* and *Chondria baileyana* (Montagne) Harvey (KU564345) for COI-5P.

The most appropriate model of sequence evolution for maximum likelihood (ML) and Bayesian inference (BI), GTR + I + G for *rbcL* and TVM + F + I + G4 for COI-5P, was selected under the Akaike information criterion (AIC) implemented in IQ-TREE webserver (Trifinopoulos et al. 2016). ML analysis was performed in IQ-TREE (Nguyen et al. 2015); nodal support was assessed with 1,000 non-parametric bootstrap (BS) replicates (Felsenstein 1985). Bayesian analysis was performed using MrBayes v3.2.2 (Ronquist et al. 2012), with four MCMC chains (one hot and three cold) for two independent runs with 4,000,000 generations and sampling one tree every 1,000 generations, starting with a random tree. We discarded the first 30,000 generations in both runs as burn-in to build the consensus tree and computing the posterior probabilities (PP). Pairwise distances were calculated using the uncorrected “*p*” distances in PAUP v4.0 beta 10 (Swofford 2002).

3 Results

3.1 Molecular analysis

A total of four new sequences were generated of *rbcL* and COI-5P, including specimens from the type localities of *Palisada corallopsis* and *P. furcata* for both markers. For *rbcL* analysis, a matrix with 76 sequences and 1,448 bp length was assembled; for COI-5P analysis, a matrix with 71 sequences and 664 bp length was assembled. The *rbcL* sequence of *P. furcata* (GU330226) previously obtained by Cassano et al. (2012b) was not included in the analysis due to its poor quality.

The *rbcL* phylogeny placed *P. furcata* from the type locality (Ceará) in the same clade with sequences of *P. corallopsis* from Cuba (type locality), Bermuda, Mexico and USA (Figure 1) with moderate to high support. The *rbcL* sequences of *P. corallopsis* presented 0.41–1.46% of intraspecific divergence, whereas *P. furcata* diverged from *P. corallopsis* in a lower range, 0.21–0.95%, with the highest value observed between sequences from their type localities (Table 1). Sequences of *P. corallopsis* and *P. furcata* were placed in a fully supported sister relationship with *P. cervicornis* from Bermuda and the USA,

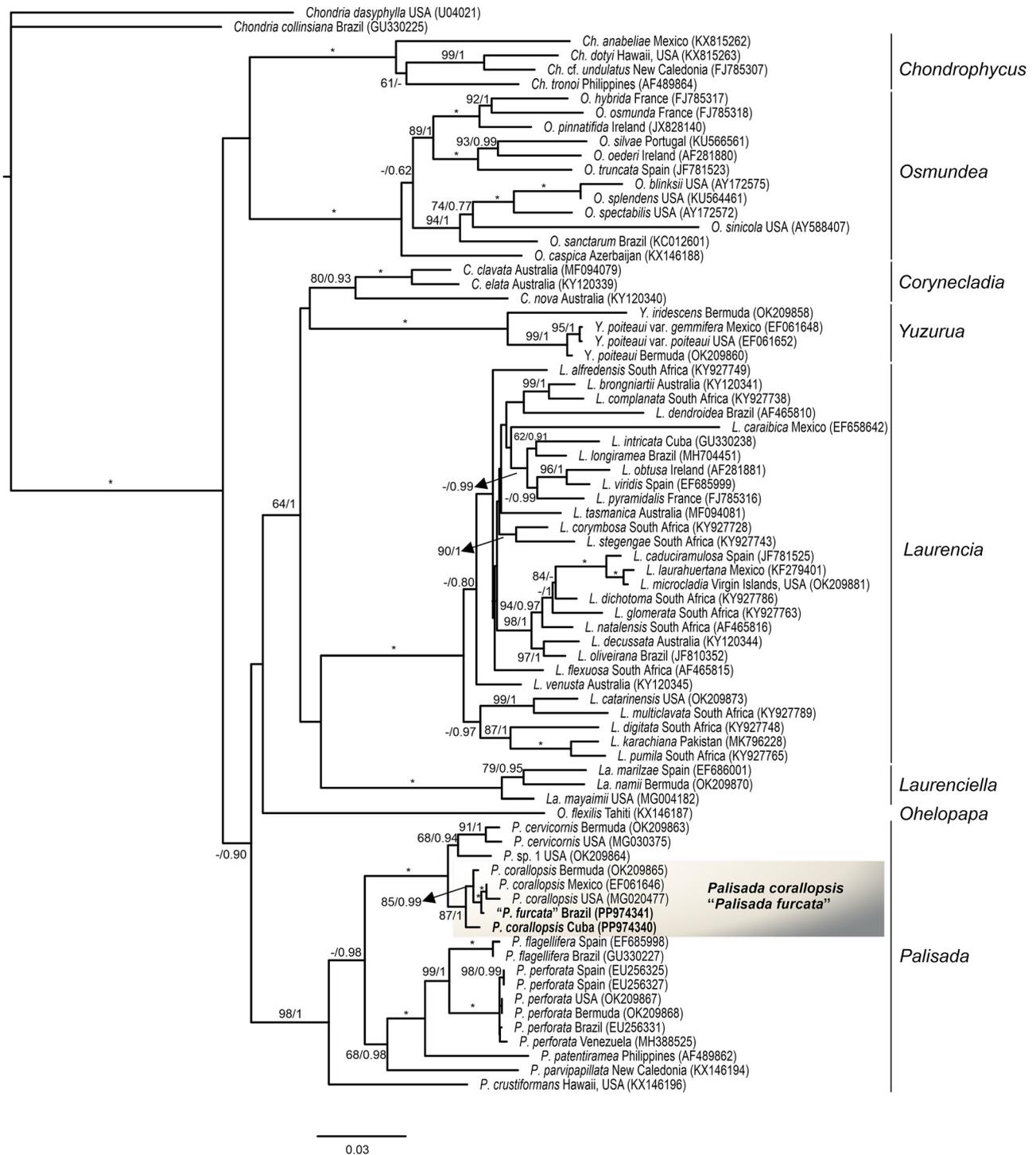


Figure 1: Maximum likelihood of *rbcL* consensus tree of the *Laurencia* complex. Only values above 60 %/0.60 for bootstrap support and Bayesian posterior probabilities are shown on branches. (-) indicates lack of bootstrap support or values under 60; (*) indicates full support. Sequences generated in this study are in bold type.

along with an unidentified species of *Palisada* (*P. sp. 1*) from Florida (Figure 1). *Palisada corallopsis*/*P. furcata* showed an interspecific divergence of 2.30–2.52 % with *P. cervicornis*.

The COI-5P results were similar to the *rbcL* with *P. furcata* from Brazil grouping with *P. corallopsis* from Cuba and Bermuda (Figure 2). The intraspecific divergence between sequences of *P. corallopsis* ranged

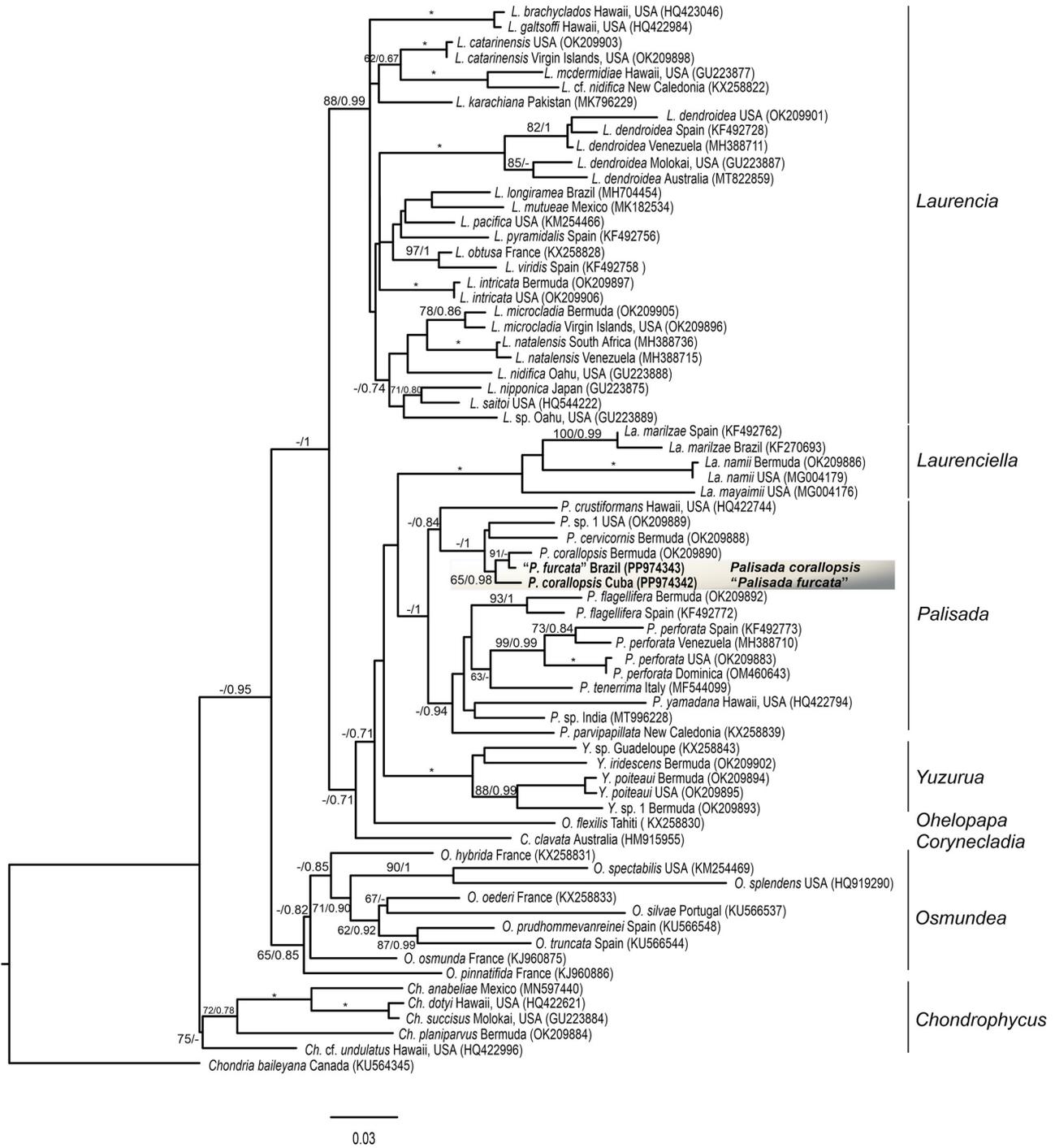


Figure 2: Maximum likelihood of COI-5P consensus tree of the *Laurencia* complex. Only values above 60 %/0.60 for bootstrap support and Bayesian posterior probabilities are shown on branches. (-) indicates lack of bootstrap support or values under 60; (*) indicates full support. Sequences generated in this study are in bold type.

1.09–2.18 %, and the sequences from the type localities of *P. furcata* and *P. corallopsis* diverged by 1.6 % (Table 1). *Palisada corallopsis*/*P. furcata* was closest to *P. cervicornis*

and also *P. sp. 1* as in the *rbcL* analyses (Figure 2). *Palisada corallopsis*/*P. furcata* diverged from *P. cervicornis* by 3.05–3.71 %.

Table 1: Comparison of the genetic divergence (%) for the molecular markers *rbcl* (above the diagonal) and COI-5P (under the diagonal) between *Palisada corallopsis* and *P. furcata*.

	<i>P. corallopsis</i> Bermuda	<i>P. corallopsis</i> Mexico	<i>P. corallopsis</i> Florida, USA	<i>P. corallopsis</i> Cuba	<i>P. furcata</i> Ceará, Brazil
<i>P. corallopsis</i> Bermuda Bermuda	–	0.57	0.98	0.82	0.49
<i>P. corallopsis</i> Mexico Mexico		–	0.41	1.02	0.21
<i>P. corallopsis</i> Florida, USA Florida, USA			–	1.46	0.6
<i>P. corallopsis</i> Cuba Cuba	2.18			–	0.95
<i>P. furcata</i> Ceará, Brazil Ceará, Brazil	1.09			1.6	–

3.2 Morphology – analysis of material from the type locality

Palisada corallopsis (Montagne) Senties, Fujii *et* Díaz-Larrea in Senties and Díaz-Larrea 2008: 69 (Figure 3 A–G, Figure 4 A–F).

Type: PC 0062369 (Herbier Muséum Paris Cryptogamie) Herb. Montagne in Paris (Yamada 1931: 198) (Figure 5).

Type locality: La Habana, Cuba.

Habit: Thalli are erect, forming large tufts, up to 8 cm high, wine-red to brownish in color, fleshy to cartilaginous in texture adhering slightly to herbarium paper

when dried (Figure 3A and B) and attached to the substratum by a large and firm discoid holdfast (Figure 3C). Thalli are cylindrical at the base, up to 3 mm in diameter and slightly flattened in the rest of the plants. Main axes are long, subdichotomously to dichotomously branched below, 0.75–2.00 mm in diameter, irregular, opposite to alternate above becoming subcorymbose. Branchlets numerous, alternate or irregular; club-shaped, short, simple, upcurved, 1–4 × 1.0–1.5 mm, usually swollen, often bifurcated, when trifurcated the branch between the bifurcations is smaller, and with slightly constricted bases (Figure 3D and E).

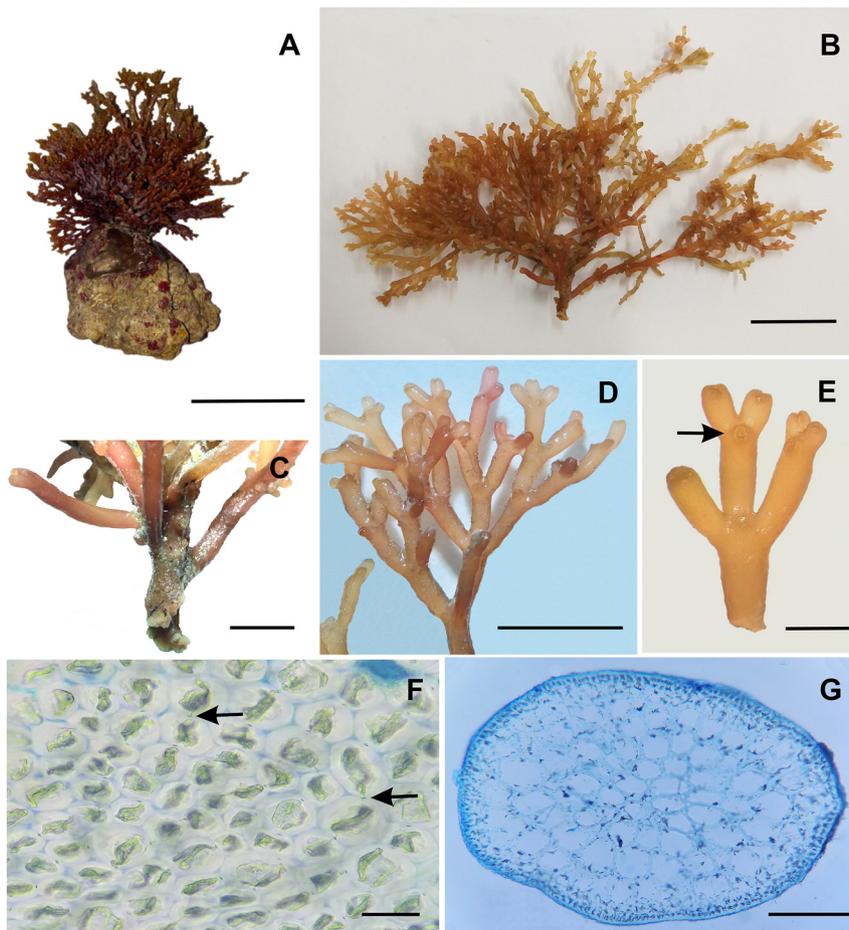


Figure 3: *Palisada corallopsis*. (A, B) Habit of the thalli. (C) Detail of basal portion of thallus. (D, E) Detail of branchlets. Note trifurcated branchlet with the smaller branchlet between the bifurcations (arrow). (F) Surface view of the cortical cells showing secondary pit-connections between the cells (arrows). (G) Transverse section of the thallus. Scale bars: A, B, 1 cm; C, D, E, 5 mm; F, 25 μ m; G, 100 μ m.

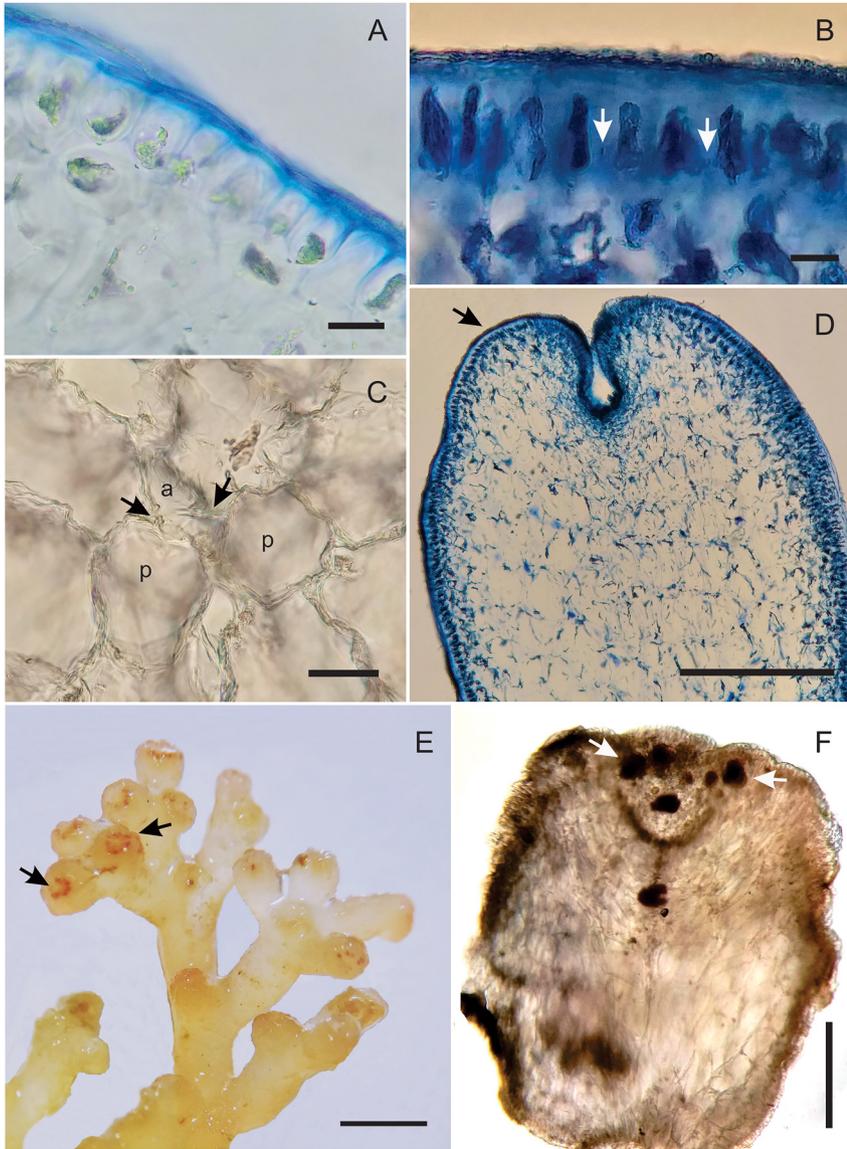


Figure 4: *Palisada corallopsis*. (A) Transverse section of a branch showing subquadratic cortical cells. (B) Longitudinal section showing cortical cells with secondary pit-connections (white arrows). (C) Transverse section of upper portion of a branch showing an axial cell (a) with two pericentral cells (p). Note pit-connections between axial and pericentral cells (arrows). (D) Longitudinal section of a branchlet. Note outer cortical cell walls near apex not projecting beyond the surface (arrow). (E) Tetrasporangial branches showing right-angle arrangement of tetrasporangia (arrows). (F) Longitudinal section through a tetrasporangial branchlet showing in detail right-angle arrangement of the tetrasporangia (white arrows). Scale bars: A, B, C, 25 µm; D, F, 100 µm; E, 3 mm.

Vegetative structures: In surface view, cells are polygonal, isodiametric, sometimes hexagonal, 23–48 µm in diameter with secondary pit-connections between adjacent cells (Figure 3F). *Corps en cerise* are absent. In transverse section of median region, thalli with 1–2 layers of pigmented cortical cells and 4–5 layers of colourless medullary cells (Figure 3G). Cortical cells are quadratic to subquadratic, rarely radially elongated and arranged as palisade-like, with some secondary pit-connection between them, 30–50 × 20–35 µm (Figure 4A and B). Medullary cells are rounded, gradually decreasing in size towards the center, thin-walled, 50–145 × 45–130 µm; pericentral cells, rounded, smaller than those of the intermediate layer. Each vegetative axial segment cuts off two pericentral cells (Figure 4C). Cell walls of the outer cortical cells at the apex not projecting above the surface of the thallus (Figure 4D). Lenticular thickening absent.

Reproductive structures: Tetrasporangial branches are swollen (Figure 4E). Tetrasporangia oval to spherical, 20–48 µm in diameter, tetrahedrally divided and arranged at right-angle in relation to the longitudinal axis of the branchlets (Figure 4E and F). No female or male plants were observed. Details of female and male reproductive structures in Cordeiro-Marino et al. (1994).

Habitat: Plants are found from the northwestern to the southeastern coast of Cuba, on rocky substrata exposed to waves, in the subtidal zone, up to 7 m deep or even in deeper waters, up to 60 m. Also found in shallow calm waters. Very abundant in coral reefs.

Specimens examined morphologically: *Palisada corallopsis* (Montagne) Senties, Fujii et Díaz-Larrea – Cuba. **La Habana.** Rincón de Guanabo, Sibarimar cove, 23° 10' 30" N, 82° 05' 48" W, 23 February 2001, leg. A.J. Areces (HANC184;



Figure 5: Herbarium sheet of the type specimen of *Sphaerococcus corallopsis* (PC0062369).

PP974340, *rbcL*; PP974342, COI-5P), 20 February 2020, leg. A.J. Areces and D. Reyes (SP514123). **Ciego de Ávila.** Cayo Coco, Playa Flamenco, 22° 31' 52" N, 78° 28' 37" W, 27 February 2008, leg. D. de la Nuez (HANC137). ***Palisada furcata*** (Cordeiro-Marino et M.T. Fujii) Cassano et M.T. Fujii – **Brazil.** **Ceará,** Trairi, Praia de Emboaca, 3° 12' 23.5" S, 39° 18' 37.1" W, 30 March 2018, leg. M.T. Fujii and L.P. Soares (SP526373; PP974341, *rbcL*; PP974343, COI-5P). **Bahia.** Salvador, Lauro de Freitas, Praia de Vilas do Atlântico, 12° 53' 38.19" S, 38° 17' 28.3" W, 24 August 2002, leg. M.T. Fujii (SP400806). **Paraíba.** Praia de Tambaú, 7° 13' 38" S, 36° 47' 23" W, 24 February 2004, leg. M.T. Fujii (SP399928), Conde, Praia de Carapibus, 7° 17' 57.01" S, 34° 47' 57.35" W, 01 April 2014, leg. M.T. Fujii (SP401518). **Pernambuco.** Jaboatão dos Guararapes, Praia de Candeias, 8° 12' 46" S, 34° 55' 6" W, 25 February 2018, leg. M. I.L.G. Cavalcanti (SP513854). **Espírito Santo.** Marataízes, Praia do Pontal, 21° 00' 19" S, 40° 48.37' W, 09 June 2017, leg. M. I.L.G. Cavalcanti (SP470199).

Additional material examined: Type: *Sphaerococcus corallopsis* Montagne (PC0062369, Figure 5). Holotype: *Laurencia furcata* Cordeiro-Marino et M.T. Fujii (SP239661, Figure 6).

4 Discussion

Palisada corallopsis was originally described by Montagne (1842) as *Sphaerococcus corallopsis* from Havana, Cuba; it

was later transferred to the genus *Laurencia* by Howe (1918). Nam (1999) transferred *Laurencia corallopsis* to the subgenus *Palisada*, section *Palisadae*, within the genus *Chondrophycus*, based on the presence of radially elongated cortical cells arranged in a palisade, with no secondary pit-connections between adjacent cells, a single sterile pericentral cell in tetrasporangial axial segments, and a right-angle arrangement of tetrasporangia. Finally, based on molecular analyses, Senties and Díaz-Larrea (2008) verified that *Chondrophycus corallopsis* (Montagne) K.W. Nam was taxonomically close to its congeners previously transferred to *Palisada* and proposed the new combination *Palisada corallopsis* (Montagne) Senties, Fujii et Díaz-Larrea.

Palisada furcata was originally proposed as *Laurencia furcata* Cordeiro-Marino et M.T. Fujii in Cordeiro-Marino et al. (1994) from Brazil. However, the authors pointed out the presence of intermediate characteristics between the subgenera *Laurencia* and *Chondrophycus* for this species, as defined by Saito (1967). These intermediate characteristics include: (i) presence of secondary pit-connections between adjacent cortical cells and (ii) cortical cells not radially elongated as palisade-like (*Laurencia*); (i) presence of two pericentral cells per vegetative axial segment, (ii) absence of *corps en cerise*, (iii) absence of lenticular thickenings, and (iv) tetrasporangia arranged at right-angles in relation to fertile branchlets (*Chondrophycus*). Subsequently, Fujii and Senties (2005) transferred *L. furcata* to *Chondrophycus*, as *C. furcatus* (Cordeiro-Marino et M.T. Fujii) M.T. Fujii et

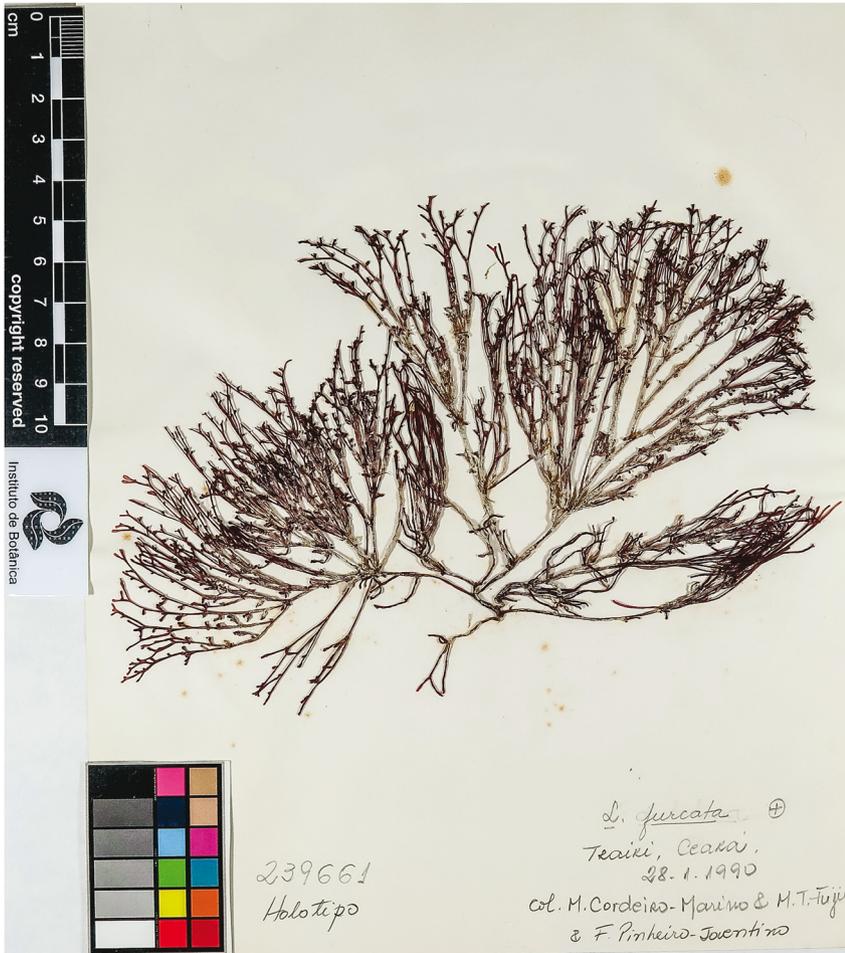


Figure 6: Herbarium sheet of the holotype of *Laurencia furcata* (SP239661).

Sentías, primarily due to the presence of two pericentral cells per vegetative axial segment. Cassano et al. (2012b), based on phylogenetic analyses, proposed the nomenclatural transfer of *C. furcatus* to *P. furcata* (Cordeiro-Marino et M.T. Fujii) Cassano et M.T. Fujii.

Our material from Cuba, collected at Rincón de Guanabo, Havana, corresponds to *Palisada corallopsis* as defined by Montagne (1842), being similar to his original description (Figure 5, Table 2). The examination of the morpho-anatomical characteristics of the type specimens (*Palisada corallopsis*, Figure 5 and *P. furcata*, Figure 6), as well as other materials of *P. furcata* from Brazil and of *P. corallopsis* from Cuba, showed that both species are very similar (Table 2). They share the presence of secondary pit-connections between cortical cells, which are radially elongated or forming a palisade-like layer, and the frequent presence of a third branch (trifurcation) at the apices of branches and branchlets. Popolizio et al. (2022) pointed out morphological differences to separate *P. furcata* from *P. corallopsis*, such as larger thallus, shape of branchlet tips (cylindrical to slightly flattened vs. swollen in *P. corallopsis*), and smaller size of the outermost cortical cells for *P. furcata*.

These characteristics can be considered intraspecific variations, as well as the diameter of the thallus of Cuban specimens, which is larger than that of Brazilian specimens (Cordeiro-Marino et al. 1994).

The distribution of *Palisada corallopsis* in the Indo-Pacific Ocean is based only on inventories without descriptions (Ang et al. 2014; Gazon-Fortes 2012; Lastimoso and Santiañez 2021; Phang et al. 2016; Silva et al. 1987, 1996), which makes it impossible for us to establish morphological comparisons. There are also no DNA sequences available in databases from this region for comparison between sequences. Thus, the citations of *P. corallopsis* from the Indo-Pacific should be reviewed through molecular data.

The *rbcL* phylogeny and low genetic divergences for both markers, 0.95 % for *rbcL* and 1.6 % for COI-5P (Table 1), strongly support the conclusion that *P. corallopsis* and *P. furcata* from the type localities correspond to the same taxonomic entity. Popolizio et al. (2022) in their *rbcL* analyses verified that *P. furcata* from Brazil (GU330226) also grouped in the *P. corallopsis* clade but showed higher divergence (1.8 %) between it and *P. corallopsis* from Mexico. The sequence of *P. furcata* (GU330226) has 14 ambiguities, which

Table 2: Comparison of morphological features between *Palisada corallopsis* and *P. furcata* from the western Atlantic Ocean; based on Popolizio et al. (2022).

Characteristics	<i>Sphaerococcus corallopsis</i>	<i>Palisada corallopsis</i>	<i>Palisada corallopsis</i>	<i>Palisada corallopsis</i>	<i>Palisada corallopsis</i>	<i>Palisada furcata</i>
Collection site/References	La Habana, Cuba (type locality)/Montagne (1842)	La Habana, Cuba (type locality)/this study	Mexican Caribbean/Senties and Fujii (2002)	Florida, USA/Dawes and Mathieson (2008)	Bermuda/Popolizio et al. (2022)	Ceará, Brazil (type locality)/Cordeiro-Marino et al. (1994), Fujii and Senties (2005)
Height plant (cm)	7.62 (= three inches)	To 8	7.5–12.0	4.0–16	To 8	To 15
Thallus shape (cross section)	–	Not cylindrical	–	–	–	Not cylindrical
Main axis basal diam. (mm)	–	3	–	–	1.5–2.0	1,050–1,560 μm
Branching pattern	Irregularly dichotomous or whorled; dichotomous above with short nearly dichotomous branchlets solitary or in clusters	Subdichotomous to ditrichotomous below; irregular, opposite to alternate above becoming subcorymbose	Spiral branching below, irregular above, up to 4 orders of branching	Subdichotomously to dichotomously branched below, irregularly alternate above	Irregularly cervicorn; apices often appearing dichotomous; some adventitious branching	Di- to trichotomous
Branchlet diam. (mm)	1–2	1.0–1.5	1–2	0.75–2.00	1–2	1.0–1.5
Branchlet tips	Slightly swollen with apical pit; trichoblasts present	Swollen; trichoblasts present	Rarely swollen	Tips are obtuse, usually swollen, with an apical cell in a terminal pit; trichoblasts present	Swollen; broad in proportion to length; trichoblasts present	–
Outer cortex cell in surface view	Hexagonal	Polygonal and isodiametric (sometimes hexagonal)	Polygonal or circular	Compact, angular to rectangular	Polygonal (hexagonal rectangular)	Polygonal and isodiametric
Arrangement of cortical cells (transverse section)	–	Quadratic to subquadratic, rarely radially elongated and arranged as palisade	Palisade	–	–	Not palisade
Outer cortex cell diam. (μm), surface view	–	23–48	8–52 \times 20–34	To 50	23–45	7–33 \times 21–33
Secondary pit-connections	–	Present	Absent	–	Present	Present
Medullary cells (μm)	–	50–145 \times 45–130	6.0–12.0	–	–	Rounded, 50–150 \times 45–135
Outer cortex cell projections	–	Absent	Absent	–	Absent	Absent
Tetrasporangium diam. (μm)	–	20–48	28–57	60–100 long	–	80–110 \times 60–75
Arrangement of tetrasporangia	–	Right-angled	Right-angled	Right-angled	–	Right-angled
Shape of spermatangial branch tips	–	–	–	–	–	Cup-shaped
Shape of cystocarp	–	–	–	Oval, subglobose to urn-shaped, and with obvious necks	–	Partly immersed

caused Popolizio et al. (2022) to raise the possibility that the distance estimate could change with better quality sequence data. In fact, the newly *rbcL* sequence of *P. furcata* diverged

by only 0.21% from Mexican sequence of *P. corallopsis* (Table 1). Furthermore, our genetic divergence results for the entire *P. corallopsis/P. furcata* clade from newly

sequences generated with good quality was slightly lower than Popolizio et al. (2022) reaching to 1.46 % for *rbcL* between *P. corallopsis* from Cuba and Florida, USA. The maximum of *rbcL* intraspecific divergence observed in our study (1.46 %) is above that found for the genus *Palisada* by Cassano et al. (2009, 0.4 %), and by Popolizio et al. (2022, 0.7 %). Nevertheless, it is below the range of interspecific variation observed between *P. corallopsis*/*P. furcata* and its closest species phylogenetically, i.e., *P. cervicornis* (2.30–2.41 % from Bermuda and 2.46–2.52 % from USA, this study; 2.7–3.5 %, Collado-Vides et al. 2017), and between *Palisada* species (2.0–3.0 %, Popolizio et al. 2022).

For COI-5P, our intraspecific divergence for *P. corallopsis*/*P. furcata* ranged from 1.09 to 2.18 %, with the higher divergence between *P. corallopsis* from Cuba and Bermuda (Table 1). This range is higher than that found by Popolizio et al. (2022, 0–1.3 %), but there was no overlap with the interspecific divergence values between *Palisada* species (Popolizio et al. 2022, 3.9–6.7 %).

In conclusion, our study provides the first DNA sequences of *P. corallopsis* and *P. furcata* from their respective type localities, Havana, Cuba, and Ceará state, Brazil, and detailed description and illustrations of the morpho-anatomical characters of the *P. corallopsis* from the type locality. Based on our morphological comparisons and molecular results, there is no evidence to consider *P. corallopsis* and *P. furcata* as different entities. Thus, *P. furcata* should be synonymized with *P. corallopsis* based on the principle of priority (ICN, Principle III, Turland et al. 2018).

5 Taxonomic change

Palisada corallopsis and *P. furcata* are here considered to be conspecific.

Palisada corallopsis (Montagne) Senties, M.T. Fujii et Díaz-Larrea in Senties and Díaz-Larrea 2008: 69.

Basionym: *Sphaerococcus corallopsis* Montagne 1842, Histoire, physique, politique et naturelle de l'île de Cuba: 49, pl. 3, fig. 1.

Homotypic synonyms: *Laurencia corallopsis* (Montagne) M. Howe 1918: 519; *Chondrophyucus corallopsis* (Montagne) K.W. Nam 1999: 463.

Heterotypic synonyms: *Laurencia furcata* Cordeiro-Marino et M.T. Fujii in Cordeiro-Marino et al. (1994: 374, figs 1–19); *Chondrophyucus furcatus* (Cordeiro-Marino et M.T. Fujii) M.T. Fujii et Senties (2005: 109, figs 69–77); *Palisada furcata* (Cordeiro-Marino et M.T. Fujii) Cassano et M.T. Fujii in Cassano et al. (2012b: 79).

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Bionotes



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